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Pongamia pinnata medicated ZnO nanoparticles preparation, characterization and anti-microbial activity

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Abstract
In the recent decades, nanotechnology has become an important research field of modern material science. Green synthesized nanoparticles have gathered wide interest in medical field due to its inherent features like rapidity, eco-friendly and cost-effectiveness. For the first time, Zinc Oxide nanoparticles were successfully synthesized using Pongamia Pinnata leaf extract in the present investigation. The optical senstivity of the Zinc Oxide nanoparticles was characterized by UV vis. The X- ray diffraction (XRD) studies showed the crystalline nature and revealed the purity of Zinc Oxide nanoparticles. The success-ability of bacteria and fungi with Synthesized ZnO nanoparticles tested using agar well diffusion method were discussed. The bacterial and fungal destruction was better for ZnO nanoparticles than reported for plant extracts and standard drugs. Thus, this study proves that Zinc Oxide nanoparticles would contain natural anti-microbial agents through green synthesis, which may produce drugs for antimicrobial therapeutics.

Keywords: Pongamia pinnata, ZnO, XRD, UV, antimicrobial activity

Introduction
In the past few years Pongamia Pinnata capped ZnO nanoparticles has drawn the attention of many researchers for their unique optical and chemical behaviours which can be easily tuned by changing the morphology. Within the large family of metal oxide, NPs ZnO have been used in various cutting edge applications like electronics, communication, sensors, cosmetics, environmental protection, biology and the medicinal industry. Moreover, ZnO has tremendous potential in biological applications like biological sensing, biological labelling, gene delivery, drug delivery and nano-medicine along with its antibacterial, antifungal, acaricidal, pediculicide, larvicidal and anti-asthmatic effect Recently, the synthesis of ZnO nanoparticles via eco-friendly routes has become popular among researchers due to its low cost, synthesis in ambient atmosphere, non-toxicity, environmental compatibility etc. and ease of applications as the resulting particles are highly soluble in water, biocompatible, and devoid of toxic stabilizers. Plant extracts are a very promising tool for the facile synthesis of ZnO nanoparticles via green routes. Citrus aurantifolia fruit juice, Parthenium hysterophorus leaf extracts, and Aloe sp. extracts have been used in ZnO synthesis by different workers. *In vivo* synthesis of ZnO nanoparticles has been also reported in Physalis alkekengi. The important properties of Pongamia Pinnata capped ZnO nanoparticles are a wide band gap of 3.36eV at room temperature and a large exciton binding energy of 60 meV which very higher than other semiconductor nano micro crystals [2].
Pongamia Pinnata is anti-asthmatic and anti-ulcer. Its has bronchodilator activity to treat respiratory disorders. Both vasicine and vasicinone the primary alkaloid constituents of Pongamia Pinnata. is also anti-inflammatory and has anti-tuberculosis activity a chemical constituent of Pongamia Pinnata alkaloids vasicine produces bromhexine and ambroxol these chemicals have ph dependent growth inhibitory effect on tuberculosis The key interest for the research for the Pongamia Pinnata capped ZnO nanoparticles nanocrystal is the high-quality material with controllable size and fine dispersion. Numerous experimental efforts have been made to fulfil the highly stable Pongamia Pinnata capped ZnO nanoparticles quantum dot with quantum confinement effect.

The optical properties of nanoparticles strongly depend on the intergranular distance and granular aggregation. Researchers have reported that aggregation of Pongamia Pinnata capped ZnO nanoparticles colloids are possible to red shifting in the absorption onset as well as photoluminescence peak [7]. In this paper, we reported that high-intensity green emission is possible in the solution synthesized Pongamia Pinnata capped ZnO nanoparticles nano microwires.

Material and Method

Collection of leaves

Pongamia pinnata, leaves were collected from Sambhar Lake Jaipur. Then washed the collected leaves with fresh water and dried under shade at room Temperature.

Preparation of extract

Dried leaves of Pongamia pinnata were break into small pieces. Then make powder by mixture blender and passed to sieve for get small and equal sized particles. Dried Powder of leaves put in clean and air tight container. Then cause the selection of solvents and mix the 20 g powder of each plant (Raut *et al*., 2014). 20 g dried mix powder was dissolve in 150 ml of ethanol, methanol and distilled water.

Green Synthesis of ZnO Nanopaarticles

Nano colloids of ZnO have been prepared by basic solution condition. The Green-ZnO quantum dot colloids were prepared from zinc acetate and NaOH with Pongamia pinnata plant extract as capping agent. For the synthesis $Zn(CH_3COO)_2.H_2O$ and NaOH were dissolved in non-hydrous ethanol at 70 °C and at room temperature respectively. 0.1 M Zn^+ solutions, 0.2M OH^- solution and Pongamia pinnata plant extract with different molar ratio were dissolved in 10ml of absolute ethanol respectively.

Microbial sample and maintenance

The gram positive bacteria *Streptococcus spp*. and gram negative bacteria *Escherichia coli* were used as test bacteria for antibacterial activity. Five fungal strains *Aspergillus niger*, *Aspergillus terreus*, *Aspergillus flavus*, *Penicillium notatum*, *Penicillium chrysogenum* were used test fungi for antifungal activity. Broth for bacteria was prepared by weighting 1.3 g of nutrient broth and then dissolving in 100ml of distilled water. Then took bacterial culture from slant with the help of inoculating loop and then inoculate bacterial culture in broth medium. Then incubate broth at 37 °C for 18-24 hours (Angelo, 2015). Broth for fungi was prepared by weighting 3.9 g of potato dextrose broth and then dissolving 100 ml of distilled water. Then took fungal culture from slant with the help of inoculating loop and then inoculate fungal culture in broth medium. Then incubate broth at 37 °C for 48 hours.

Preparation of Media

Bacterial media: Nutrient agar media was used for the growth of two bacterial strains.

Fungal media: Sabouraud dextrose agar (SDA) media was used for the growth of five fungal strains.

Antibacterial assay

Disc diffusion method was used to determine the antibacterial activity of aqueous Green ZnO extract and solvent Green ZnO extracts (Methanol and ethanol) on nutrient agar media. The 2.8 g nutrient agar weighted and dissolved in 100 ml of distilled water in a clean conical flask. Then autoclaving the media and cool at room temperature. Poured media into sterile petriplates, Bacterial culture was spread on the agar plates using a sterile cotton swab for obtain regular microbial Growth [13].

Results

Structural analysis (XRD)

The XRD results accompanied with Rietveld analysis indicate that the final ZnO is mainly oxide of zinc, whereas the intermediate is mainly sulphide of zinc. The dotted curve in Fig 1 represents a typical observed XRD pattern from the capped sample. All the observed diffraction peaks correlated well with the wurtzite structure of ZnO with space group P63mc (JCPDS File No 186). XRD pattern of uncapped sample S1 exhibited 7 peaks positioned at 2θ= 31.66°, 34.40°, 36.14°, 47.50°, 56.56°, 62.76° and 67.62° corresponding to (100), (002), (101), (102), (110), (103) and (113) planes of wurtzite phase of ZnO. The absence of any other chemical phase indicated purity and crystallinity of as prepared ZnO sample. Fig 1 shows the XRD pattern Pongamia Pinnata capped ZnO nanoparticles synthesised by above outlined procedure. All the diffraction peaks of samples adopt hexagonal wurtzite structure with phase group P63mc (186). There were no diffraction peak noticed corresponding to Zn $(OH)_2$ or any other impurities. Particle sizes of the Jasada Bhasam samples were calculated from the broadening of the different peaks by using Shrerrer formula. The average diameter of the particle was calculated as 25 nm for sample respectively where the Bohr exciton radius of Pongamia Pinnata capped ZnO nanoparticles is 2.3 nm [8].

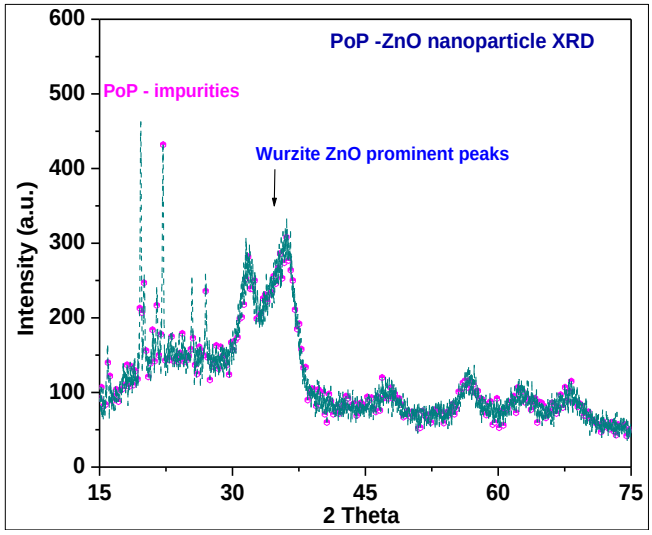

Fig 1: XRD spectra Pongamia Pinnata capped ZnO nanoparticles sample

Optical characterization

Upper part of Fig 2 shows UV-vis spectra for the synthesized Pongamia Pinnata capped ZnO nanoparticles, prepared from Zinc acetate with wavelength ranging from 280-600 nm. The spectra of Pongamia Pinnata capped ZnO nanoparticles microwires different ratio of Pongamia Pinnata extract capping agent dispersed in ethanol, exhibit well cleared exciton peak and absorption onset. Absorption onset of sample has onset at 360 in the absorption spectra

shows an obvious blue shift from bulk Pongamia Pinnata capped ZnO nanoparticles, which were shown at 382 nm. All the absorption characterization has been carried out one hour after the synthesis [9]. Photo luminescence properties of Pongamia Pinnata capped ZnO nanoparticles dispersed in ethanol was also investigated in Fig 2. All the spectra have two emission peaks. One is relatively weak UV emission peak in 360-380 range, which is near to absorption onset, is interpreted as band edge emission. Second is a broad trap emission peak, maxima is ranging from 520 to 535 which is in green luminescence region. This broadness of trap emission is ranging from 440 to 700 nm. Both the band edge emission peaks and trap emission peaks are shifted towards the lower energy side corresponding to absorption shift [10].

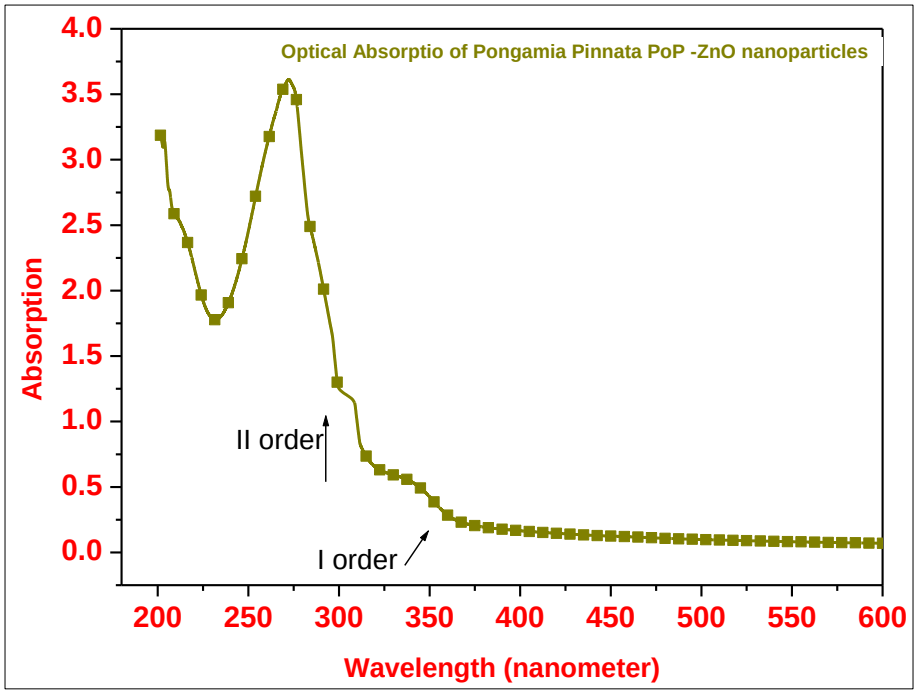

Fig 2: Optical absorption spectra Pongamia Pinnata capped ZnO nanoparticles sample

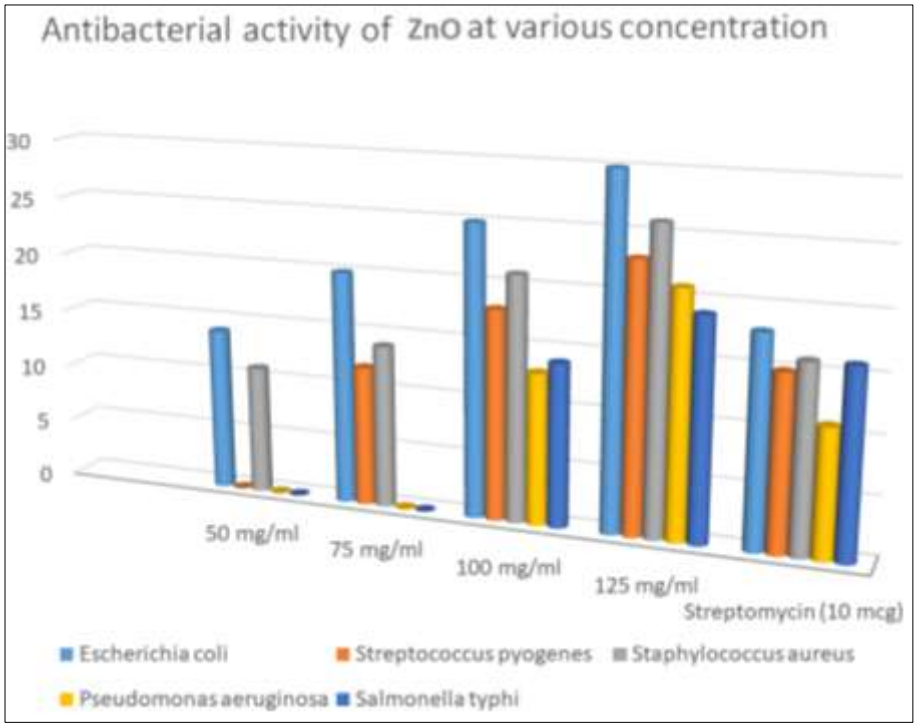

Fig 3: Antimicrobial activity of Pongamia Pinnata capped ZnO nanoparticles sample

The goal of the current investigation was to evaluate the synthesized Pongamia Pinnata capped ZnO nanoparticles' antibacterial activity. The antibacterial activity of the study's findings was measured and recorded based on the diameter of the zone of inhibition. Pongamia Pinnata capped ZnO nanoparticles were produced at various doses to examine their antibacterial properties. The outcomes were contrasted with the antimicrobial medication Streptomycin, which is sold commercially. According to the study, pongamia pinnata medicated or Pongamia Pinnata capped ZnO nanoparticles shown strong antibacterial action against *Salmonella typhi*, *Pseudomonas aeruginosa*, *Staphylococcus*

aureus, Escherichia coli, and Streptococcus pyogenes. Pongamia Pinnata capped ZnO nanoparticles at 50 mg/ml concentration suppressed Salmonella typhi, Pseudomonas aeruginosa, and Streptococcus pyogenes growth, but not that of Escherichia coli or Staphylococcus aureus. Pongamia Pinnata capped ZnO nanoparticles exhibited excellent antibacterial action, with the greatest zone of inhibition up to being found. In the research article, the best combination to inhibit the growth of bacteria was found to be a 125 mg/ml concentration of green synthesized Pongamia Pinnata capped ZnO nanoparticles. The results of the present study indicate that prepared Pongamia Pinnata capped ZnO nanoparticles was equally effective as the standard antimicrobial drug Streptomycin [11].

Table 1: Antibacterial activity of green synthesized Pongamia Pinnata capped ZnO nanoparticles sample

The concentration of ZnO/Standard	Zone of Inhibition				
	Escherichia coli	***Streptococcus pyogenes***	***Staphylococcus aureus***	***Pseudomonas aeruginosa***	***Salmonella typhi***
50 mg/ml	13	ND	10	ND	ND
75 mg/ml	21	13	15	ND	ND
100 mg/ml	26	19	20	14	15
125 mg/ml	31	23	25	18	20
Streptomycin (10mcg)	19	16	15	10	17

Discussion

The synthesized fresh Pongamia Pinnata capped ZnO samples shows the smaller cluster size using SEM and lowest cluster size of the Pongamia Pinnata capped ZnO nanoparticles quantum dots is 25 nm. We can understand that the suggested technique for creating nanocrystals is regarded as a feasible and environmentally safe method. Using XRD, UV, respectively, the structural, morphological & chemical analyses of green - eco-friendly synthesized pongamia pinnata medicated ZnO nanoparticles were conducted. It is well established that pongamia pinnata medicated ZnO nanoparticles can be utilized for the antifungal and antibacterial studies. From the antimicrobial studies it is inferred that surface of metal nanoparticles containing plant extracts exhibit excellent bacteriostatic effect against different microorganism. The combined form of ayurvedic plant extract & ZnO nano medicine containing capped and charged metal nanoparticles show an enhanced antimicrobial effect very deeply. Hence we can use the nanocomposite or plant extract nanomedicine to cure microbial resistant microorganism [10-12].

Conclusion

In conclusion Pongamia Pinnata capped ZnO nanoparticles were successfully produced at room temperature via a green synthesis process. The UV and XRD results show the high quality of the Pongamia Pinnata capped ZnO nanoparticles crystal structure. Good optical properties also shown in the results of Pongamia Pinnata capped ZnO nanoparticles at 336 nm. Pongamia Pinnata extract and Pongamia Pinnata capped ZnO nanoparticles were prepared in various concentrations to examine its antibacterial activity. The outcomes were contrasted with the antibacterial medication Streptomycin, which is sold commercially. According to the study, Salmonella typhi, & Escherichia coli, were all successfully combatted by nanoparticles formulations. *E. coli* and *S. typhi* growth was inhibited by Pongamia Pinnata capped ZnO at a concentration of 30 mg/ml. According to this study, 30 mg/ml of Pongamia Pinnata capped ZnO nanoparticles was the optimal concentration to prevent bacterial growth.

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